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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
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EXAMINER	
SHEINBERG, MONIKA B	

ART UNIT	PAPER NUMBER
1634	

DATE MAILED: 02/11/2004

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	09/832,621	RAUCY, JUDY	
	Examiner	Art Unit	
	Monika B Sheinberg	1634	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *27 October 2003*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *84-149* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *84-149* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. §§ 119 and 120

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

13)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application) since a specific reference was included in the first sentence of the specification or in an Application Data Sheet. 37 CFR 1.78.

a) ☐ The translation of the foreign language provisional application has been received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121 since a specific reference was included in the first sentence of the specification or in an Application Data Sheet. 37 CFR 1.78.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449) ~~Paper No(s).~~ *3 sheets*.
4) ☐ Interview Summary (PTO-413) Paper No(s). _____.
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☒ Other: *Detailed Action*.

DETAILED ACTION

Response to Amendment Filed: 27 October 2003

1. Applicants' arguments, filed: 27 October 2003, have been fully considered but they are not deemed to be persuasive. Rejections and/or objections not reiterated from previous office actions are hereby withdrawn. The following rejections and/or objections are either reiterated or newly applied. They constitute the complete set presently being applied to the instant application.

2. The cancellation of claims 21-83 is acknowledged. The addition of new claims 84-149 is acknowledged and entered.

3. Claims 84-149 are hereby examined.

-MAINTAINED REJECTIONS-

Claim Rejections - 35 USC § 102

4. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

5. Claims 116, 118, 119, 126-131, 133-137, 139-143, 145 and 146 are rejected under 35 U.S.C. 102(b) as being anticipated by Honkakoski et al. (*Mol. Cell. Biol.*, Oct. 1998). This rejection is reiterated and maintained with respect to the corresponding canceled rejected claims 52-54, 61-66, 68-72, 74-76, 79 and 80.

> The instant claim 116 requires the following: a method for screening compounds for those that alter the desired protein's expression, thereby indicating some modification in drug metabolism, comprising a test compound and a cell comprising:

- A first nucleic acid molecule comprising: a native promoter or enhancer operable for a nucleic acid molecule encoding a protein involved in drug metabolism; and operable for a reporter gene
- A second nucleic acid molecule encoding an intracellular receptor or transcription factor that upon ligand (i.e. drugs) interaction will directly or indirectly activate the promoter and in turn the desired protein and reporter gene.

Honkakoski et al. demonstrates an isolated and cultured (*claims 142, 143*) murine hepatocytic cell line (*claims 141, 145, 146*) and an up-scalable method (*claim 118*) of screening xenobiotics for induced cytochrome P-450 protein (CYP)(*claim 119*) expression as per the required limitations of the claims. The cell and method utilize an endogenous enhancer that is native to the CYP2B genes and within the chromosome (Phenobarbital-responsive enhancer module: PBREM) (*claims 116, 119, 129, 131*). The second nucleic acid comprises the transfected intracellular receptors CAR and retinoid X receptor (RXR) which upon ligand interaction bind to specific binding sites on the PBREM for activating the expression of the desired gene and PBREM linked reporter genes such as CAT reporter plasmid (p. 5652, 1st column, 2nd paragraph to p. 5653, 1st column, 3rd paragraph) and the betagalactosidase plasmid (figure 7) (*claims 116, 126-130*). The transfected receptors are indicated to be endogenously expressed therefore also within the chromosome of the cell (p. 5656, 1st column, lines 1-2) (*claims 139, 140*). The reference indicates that the orphan receptors (*claims 136*) identified are suggested to "provide cells with the capability to induce the various CYP genes and other genes responsive to unlimited numbers of xenobiotic chemicals" (p. 5657, bridging paragraph between columns, last line). The reference also suggests that the intracellular receptors or transcription factors could be hormone receptors (p. 5657, 1st column, last paragraph) (*claims 137*). The drug, chemical, and metabolite limitations of claims 133-135 are encompassed by the statement of the reference "[t]he CYP-dependent metabolism can [...] produce a practically unlimited number of potential ligands (both endogenous hormones and exogenous chemicals) for the nuclear receptors" (p. 5657, 1st column, last paragraph). Therefore Honkakoski et al. anticipates the instant claims.

Response to Arguments

6. Applicants argue that Honkakoski "does not teach recombinant cells stably transfected with one or both of the recited nucleic acid molecules". This argument that that Honkakoski does not teach that either of the nucleic acid molecules are stably transfected (interpreted to be stably incorporated) but instead teaches transient transfections is found persuasive with respects to those claims that require one or both of the nucleic acids to be stably transfected (as interpreted to be 'incorporated' into the chromosome of the cell). Therefore the rejection has been withdrawn from claims amended to require the stable transfection of one or both of the nucleic acids. However this limitation is not a requirement of all the claims such as claims 116, 118, 119, 126-131, 133-137, 139-143, 145 and 146. As such, with respect to the instant claims 116, 118, 119, 126-131, 133-137, 139-143, 145 and 146; this argument is not found to be persuasive.

7. Applicant further argues that Honkakoski performs a method rather "*to identify orphan receptors* (CAR, RXR) *that can bind the promoter or enhancer fragments* to regulate gene expression in response to a particular modulating compound" than to evaluate compounds. This argument is not found persuasive because the method of the claim comprises a test compound and a recombinant cell in which the both elements and limitations within are taught by the reference: the recombinant cell of the reference still comprises the limitations of the claim; the orphan receptors are involved in drug metabolism; and the test compounds are screened for their ability to induce gene expression. Thus Honkakoski teaches each element and method step of the claims. The intended use stated in the preamble of the claim is not sufficient to distinguish the method steps of the claim from the teachings of Honkakoski.

As such the rejection is maintained with respect to claims 116, 118, 119, 126-131, 133-137, 139-143, 145 and 146.

Claim Rejections - 35 USC § 103

8. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

9. Claims 120-125, 144 and 147-149 are rejected under 35 U.S.C. 103(a) as being unpatentable over Honkakoski et al. (*Mol. Cell. Biol.*, Oct. 1998) in view of Iyer et al. (*Europ. J. Cancer*, 1998) and Windmill et al. (*Mutation Research*, 1997). This rejection is reiterated and maintained with respect to the corresponding canceled rejected claims 55-60, 78 and 81-83.

The instant claim 116 requires the following: a method for screening compounds for those that alter the desired protein's expression, thereby indicating some modification in drug metabolism, comprising a test compound and a cell comprising:

- A first nucleic acid molecule comprising: a native promoter or enhancer operable for a nucleic acid molecule encoding a protein involved in drug metabolism; and operable for a reporter gene
- A second nucleic acid molecule encoding an intracellular receptor or transcription factor that upon ligand (i.e. drugs) interaction will directly or indirectly activate the promoter and in turn the desired protein and reporter gene.

Honkakoski et al. demonstrates an isolated and cultured murine hepatocytic cell line and an up-scalable method of screening xenobiotics for induced cytochrome P-450 protein (CYP) expression as per the required limitations of the claims. The cell and method utilize an endogenous enhancer that is native to the CYP2B genes and within the chromosome (Phenobarbital-responsive enhancer module: PBREM). The second nucleic acid comprises the transfected intracellular receptors CAR and retinoid X receptor (RXR) which upon ligand interaction bind to specific binding sites on the PBREM for activating the expression of the desired gene and PBREM linked reporter genes such as CAT reporter plasmid (p. 5652, 1st column, 2nd paragraph to p. 5653, 1st column, 3rd paragraph) and the betagalactosidase plasmid (figure 7). The transfected receptors are indicated to be endogenously expressed therefore also within the chromosome of the cell (p. 5656, 1st column, lines 1-2). The reference indicates that the orphan receptors identified are suggested to "provide cells with the capability to induce the various CYP genes and other

genes responsive to unlimited numbers of xenobiotic chemicals" (p. 5657, bridging paragraph between columns, last line). The reference also suggests that the intracellular receptors or transcription factors could be hormone receptors (p. 5657, 1st column, last paragraph). The drug, chemical, and metabolite limitations are encompassed by the statement of the reference "[t]he CYP-dependent metabolism can [...] produce a practically unlimited number of potential ligands (both endogenous hormones and exogenous chemicals) for the nuclear receptors" (p. 5657, 1st column, last paragraph).

Honkakoski et al. does not teach the various other drug metabolizing enzymes of claims 120-125, nor the various tissues of claims 144, 147-149.

Iyer et al. is a teaches the use of pharmacogenetics in screening drug toxicity that evaluate specific drug metabolizing enzymes (*claims 119-125*) including glutathione S-thansferases, uridine diphosphate glucuronosyl-transferases, and cytochrome P450 enzymes (abstract, lines 5-8).

Windmill et al. demonstrates the expression of various drug metabolizing enzymes in a multitude of human organ systems (*claims 144, 147-149*). For example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung"; "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16); and a cytochrome P450 expression in "human liver, stomach, small and large intestine, gall bladder, appendix, lung, kidney and adrenals (p.156, 1st column, 1st paragraph).

10. Thus, it would have been obvious *prima facia* to one of ordinary skill in the art at the time of the invention was made to perform the method of screening xenobiotics of Honkakoski et al and further modify the protein involved in drug metabolism to include other proteins that contribute to drug metabolism such as in chemoresistance such as glucuronosyl transferases, N-acetyltransgerases, p-glyoproteins, gluthathione transgerases and sulfotransferases as per the teachings of Iyer et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Iyer et al. in the instant screening method because of the advantages of studying the expression of the instant drug metabolizing enzymes which play a key role in cancer therapy. In addition, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to further modify the cells of the method to included those of an isolate

human cell line and or various systems other than the liver (lung, gastrointestinal tract and kidney) as per the teachings of Windmill et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Windmill et al. in the instant screening method due to these drug metabolizing enzymes are not limited to one organ system: for example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung" and "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16).

Response to Arguments

11. Applicant's arguments are directed to the reference Honkakoski. These arguments have been thoroughly reviewed but were not found persuasive for reasons already made of record in section numbers 4 and 5 above.

-NEW GROUNDS OF REJECTION-

Claim Rejections - 35 USC § 102

12. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

13. Claims 84, 85, 92, 93, 95-101, 105-112, 116, 117, 119, 120. 126, 127, 129-135 and 139-146 are rejected, as necessitated by amendment, under 35 U.S.C. 102(b) as being anticipated by Quattrochi et al. (*Mol. Pharmacol.*, 1993).

Quattrochi teaches stable cytochrome P450 expression systems (*claims 85, 119*) that transform a human (*claims 107, 110, 141, 144*) hepatoma cell line (*claims 111, 112, 145, 146*) to perform a method of testing the toxicological effect of various concentrations of compounds (*claims 100, 134*) that are utilized as drugs (*claims 99, 133*) in gastroesophageal reflux disease

therapy (abstract and p. 504, 2nd column, 2nd paragraph) (*claims 84, 116, 117*). The cultured recombinant cells (thus also isolated)(*claims 108, 109, 142, 143*) comprise:

- a first nucleic acid with a human CYP1A1 promoter (a native promoter and also an endogenous promoter)(*claim 84, 97, 116, 131*) fused to a firefly luciferase gene (reporter gene)(*claims 92, 93, 126, 127*); and
- a second nucleic acid that is endogenous to the cell (thus within the chromosome of the cell)(*claims 105, 106, 139, 140*) that encodes an Ah receptor, a transcription factor, which induces the expression of the protein involved in drug metabolism (CYP1A1),

as required by claims 84 and 116 (p. 505, 1st column, 3rd paragraph to 2nd column, last paragraph). The first nucleic acid and the reporter gene are stably incorporated (p. 505, 2nd column, last paragraph). [therefore becoming within the chromosome of the cell and endogenous (*claims 95-98, 129-132*)]. Quattrochi contemplates that a metabolite of the compound (*claim 101, 135*) could be the induction agent of the CYP genes (p. 507, 1st column, last paragraph).

14. Claims 116, 118, 119, 126-128, 131, 133-143, 145 and 149 are rejected, as necessitated by amendment, under 35 U.S.C. 102(b) as being anticipated by Kliewer et al. (*Cell*, 1998; recent IDS).

Kliewer at al demonstrates a screening assay for the induction pregnane X receptor (PXR, a hormone orphan receptor)(*claims 136, 137*) in a transiently transfected isolated mammalian kidney cell line (CV-1; p. 75, 2nd column, 1st paragraph) (*claims 141, 142, 145, 149*). The transfected CV-1 cells "were systematically treated with a series of natural and synthetic compounds that included steroids, vitamin D analogs, thyroid hormone analogs, retinoids, fatty acids, and other small, lipophilic molecules, and reporter levels measured" (p. 75, 2nd column, 1st paragraph) (*claims 116, 118, 133-135*). The cultured recombinant cells (*claim 143*) compromise:

- an extrachromosomal first nucleic acid (*claim 128*) with a human CYP3A1 promoter (a native promoter and also an endogenous promoter) (*claim 116, 131*) a CAT reporter gene (*claims 126, 127*); and
- an extrachromosomal second nucleic acid (*claim 138*) that is also endogenous to the cell (thus within the chromosome of the cell)(*claims 139, 140*) that encodes an PXR

which induces the expression of cytochrome P450 proteins involved in drug metabolism (CYP3A1) (*claim 119*),

as required by claim 116 (p. 80, 1st to 2nd column, *Experimental Procedures*). As such Kliewer et al teaches the limitations of the instant claims 116, 118, 119, 126-128, 131, 133-143, 145 and 149.

Claim Rejections - 35 USC § 103

15. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The instant claim 116 requires the following: a method for screening compounds for those that alter the desired protein's expression, thereby indicating some modification in drug metabolism, comprising a test compound and a cell comprising:

- A first nucleic acid molecule comprising: a native promoter or enhancer operable for a nucleic acid molecule encoding a protein involved in drug metabolism; and operable for a reporter gene
- A second nucleic acid molecule encoding an intracellular receptor or transcription factor that upon ligand (i.e. drugs) interaction will directly or indirectly activate the promoter and in turn the desired protein and reporter gene.

16. Claims 84-115, 117, 128-130, 132, 138 and 139 are rejected, as necessitated by amendment, under 35 U.S.C. 103(a) as being unpatentable over Honkakoski et al. (*Mol. Cell. Biol.*, Oct. 1998) in view of Iyer et al. (*Europ. J. Cancer*, 1998) and Windmill et al. (*Mutation Research*, 1997); and further in view of Makrides (*Protein Expr. Purif.*, 1999).

Honkakoski et al. demonstrates an isolated and cultured (*claims 108, 109*) murine hepatocytic cell line (*claims 107, 111, 112*) and an up-scalable method of screening xenobiotics for induced cytochrome P-450 protein (CYP) (*claim 85*) expression as per the required limitations of the claims. The cell and method utilize an endogenous enhancer that is native to

the CYP2B genes and within the chromosome (Phenobarbital-responsive enhancer module: PBREM) (*claims 84, 85,95, 97*). The second nucleic acid comprises the transfected intracellular receptors CAR and retinoid X receptor (RXR) which upon ligand interaction bind to specific binding sites on the PBREM for activating the expression of the desired gene and PBREM linked reporter genes such as CAT reporter plasmid (p. 5652, 1st column, 2nd paragraph to p. 5653, 1st column, 3rd paragraph) and the betagalactosidase plasmid (figure 7) (*claims 84, 92-96*). The transfected receptors are indicated to be endogenously expressed therefore also within the chromosome of the cell (p. 5656, 1st column, lines 1-2) (*claims 105, 106*). The reference indicates that the orphan receptors (*claims 102*) identified are suggested to "provide cells with the capability to induce the various CYP genes and other genes responsive to unlimited numbers of xenobiotic chemicals" (p. 5657, bridging paragraph between columns, last line). The reference also suggests that the intracellular receptors or transcription factors could be hormone receptors (p. 5657, 1st column, last paragraph) (*claims 103*). The drug, chemical, and metabolite limitations of claims 99-101 are encompassed by the statement of the reference "[t]he CYP-dependent metabolism can [...] produce a practically unlimited number of potential ligands (both endogenous hormones and exogenous chemicals) for the nuclear receptors" (p. 5657, 1st column, last paragraph).

Honkakoski et al. does not teach the various other drug metabolizing enzymes of claims 86-91, nor the various tissues of claims 110, 113-115 while utilizing a recombinant cell that has the first **and/or** second nucleic acid molecule stably transfected into said cell as required by claims 84 and 117.

Iyer et al. is a teaches the use of pharmacogenetics in screening drug toxicity that evaluate specific drug metabolizing enzymes (*claims 86-91*) including glutathione S-thansferases, uridine diphosphate glucuronosyl-transferases, and cytochrome P450 enzymes (abstract, lines 5-8).

Windmill et al. demonstrates the expression of various drug metabolizing enzymes in a multitude of human organ systems (*claims 110, 113-115*). For example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung"; "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16); and a cytochrome P450 expression in "human liver,

stomach, small and large intestine, gall bladder, appendix, lung, kidney and adrenals (p.156, 1st column, 1st paragraph).

Makrides teaches the advantages in developing mammalian gene expression vectors that can be successfully transiently or stably transfected for a broad range of cell types (abstract). While Makrides teaches that with respect to transient transfection, "the method lends itself to the rapid testing of vector functionality as well as optimization of different combinations of promoters and other elements in expression vectors" (p.184, 2nd column, 1st paragraph). Makrides also teaches that stable transfections (*claims 84, 117*) are also desirable because the gene expression within the cell lines would be 'permanent' (p. 185, 1st column, 2nd paragraph). In addition, Makrides states that stable transfections can be either a "stable integration of plasmid into the host chromosome [(*claims 95, 96, 105, 129, 130, 139*); it is to be noted, once a cell is stably transfected the transfected nucleic acids become endogenous to the progeny (*claim 98, 132*); OR] it is also possible, however, to generate stable cell lines that harbor vectors extrachromosomally [(*claims 94, 128, 138*)]" (p. 185, 1st column, 2nd paragraph).

17. Thus, it would have been *prima facia* obvious to one of ordinary skill in the art at the time of the invention was made to perform the method of screening xenobiotics and make the recombinant cell of Honkakoski et al, and further modify the protein involved in drug metabolism to include other proteins that contribute to drug metabolism such as in chemoresistance such as glucuronosyl transferases, N-acetyltransgerases, p-glyoproteins, gluthathione transgerases and sulfotransferases as per the teachings of Iyer et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Iyer et al. in the instant recombinant cell and screening method due to the advantages of studying expression of the instant drug metabolizing enzymes play a key role in cancer therapy. In addition, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to further modify the cells of the method to included those of an isolate human cell line and or various systems other than the liver (lung, gastrointestinal tract and kidney) as per the teachings of Windmill et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Windmill et al. in the instant recombinant cell and screening method due to these drug metabolizing enzymes are not limited to one organ system: for example N-acetyltransferase "mRNA expression was detected in human liver, small

intestine, colon, esophagus, bladder, ureter, stomach and lung" and "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16). Furthermore, it would have been *prima facia* obvious to one of ordinary skill in the art at the time of the invention was made to perform the method of Honkakoski in view of Iyer et al. and Windmill et al.; and further stably transfect at least one of the nucleic acids as per the teaching of Makrides. One of ordinary skill in the art would have been motivated to modify the method and recombinant cell because the alternative embodiments allow for greater versatility in the utility of the method and recombinant cell. The nucleic acids can exist stably in various capacities wherein the 1st and/or 2nd nucleic acids can be either stably incorporated into the genome of the cell **or** be stably transfected while remaining as an extrachromosomal element as per the teachings of Makrides. In addition, an ordinary artisan would have been further motivated to perform the modified method with the first and/or the second nucleic acid molecules stably transfected due to the advantages to having a cell line for a screening assay that permanently expresses the transfected construct instead a cell line with limited timeline of expression as seen in transient transfections.

18. Claims 86-91, 113-115, 120-125 and 147-149 are rejected, as necessitated by amendment, under 35 U.S.C. 103(a) as being unpatentable over Quattrochi et al. (*Mol. Pharmacol.*, 1993) in view of Iyer et al. (*Europ. J. Cancer*, 1998) and Windmill et al. (*Mutation Research*, 1997).

Quattrochi teaches stable cytochrome P450 expression systems that transform a human hepatoma cell line to perform a method of testing the toxicological effect of various concentrations of compounds that are utilized a drug in gastroesophageal reflux disease therapy (abstract and p. 504, 2nd column, 2nd paragraph). The cultured recombinant cells (thus also isolated) comprise:

- a first nucleic acid with a human CYP1A1 promoter (a native promoter and also an endogenous promoter) fused to a firefly luciferase gene (reporter gene); and
- a second nucleic acid that is endogenous to the cell (thus within the chromosome of the cell) that has the Ah receptor, a transcription factor, which induces the expression of

the protein involved in drug metabolism (CYP1A1). (p. 505, 1st column, 3rd paragraph to 2nd column, last paragraph).

The first nucleic acid and the reporter gene are stably incorporated, therefore becoming within the chromosome of the cell and endogenous (p. 505, 2nd column, last paragraph). Quattrochi contemplates that a metabolite of the compound could be the induction agent of the CYP genes (p. 507, 1st column, last paragraph).

Quattrochi does not teach the various other drug metabolizing enzymes of claims 86-91 and 120-125, nor the various tissues of claims 113-115 and 147-149.

Iyer et al. is a teaches the use of pharmacogenetics in screening drug toxicity that evaluate specific drug metabolizing enzymes (*claims 86-91, 120-125*) including glutathione S-thansferases, uridine diphosphate glucuronosyl-transferases, and cytochrome P450 enzymes (abstract, lines 5-8).

Windmill et al. demonstrates the expression of various drug metabolizing enzymes in a multitude of human organ systems (*claims 113-115, 147-149*). For example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung"; "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16); and a cytochrome P450 expression in "human liver, stomach, small and large intestine, gall bladder, appendix, lung, kidney and adrenals (p.156, 1st column, 1st paragraph).

19. Thus, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to perform the method of toxicological testing and make the recombinant cell of Quattrochi, and further modify the protein involved in drug metabolism to include other proteins that contribute to drug metabolism such as in chemoresistance such as glucuronosyl transferases, N-acetyltransgerases, p-glyoproteins, gluthathione transgerases and sulfotransferases as per the teachings of Iyer et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Iyer et al. in the instant recombinant cell and screening method because the advantages of studying expression of the instant drug metabolizing enzymes play a key role in cancer therapy. In addition, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to further modify the cells of the method to included those of an isolate human cell line and or various systems other than

the liver (lung, gastrointestinal tract and kidney) as per the teachings of Windmill et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Windmill et al. in the instant recombinant cell and screening method because these drug metabolizing enzymes are not limited to one organ system: for example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung" and "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16).\

20. Claims 120-125, 144 and 146-148 are rejected, under 35 U.S.C. 103(a) as being unpatentable over Kliewer et al. (*Cell*, 1998; recent IDS) in view of Iyer et al. (*Europ. J. Cancer*, 1998) and Windmill et al. (*Mutation Research*, 1997).

Kliewer at al demonstrates a screening assay for the induction pregnane X receptor (PXR, a hormone orphan receptor) in transiently transfected isolated mammalian kidney cell line (CV-1; p. 75, 2nd column, 1st paragraph). The transfected CV-1 cells "were systematically treated with a series of natural and synthetic compounds that included steroids, vitamin D analogs, thyroid hormone analogs, retinoids, fatty acids, and other small, lipophilic molecules, and reporter levels measured" (p. 75, 2nd column, 1st paragraph). The cultured recombinant cells compromise:

- an extrachromosomal first nucleic acid with a human CYP3A1 promoter (a native promoter and also an endogenous promoter) a CAT reporter gene; and
- an extrachromosomal second nucleic acid that is also endogenous to the cell (thus within the chromosome of the cell) that encodes an PXR which induces the expression of cytochrome P450 proteins involved in drug metabolism (CYP3A1),

as required by claim 116 (p. 80, 1st to 2nd column, *Experimental Procedures*). Kliewer et al does not teach the various other drug metabolizing enzymes of claims 120-125, nor the various tissues of claims 146-148.

Iyer et al. is a teaches the use of pharmacogenetics in screening drug toxicity that evaluate specific drug metabolizing enzymes (*claims 120-125*) including glutathione S-thansferases, uridine diphosphate glucuronosyl-transferases, and cytochrome P450 enzymes (abstract, lines 5-8).

Windmill et al. demonstrates the expression of various drug metabolizing enzymes (*claims 146-148*) in a multitude of human (*claims 144*) organ systems. For example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung"; "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16); and a cytochrome P450 expression in "human liver, stomach, small and large intestine, gall bladder, appendix, lung, kidney and adrenals (p.156, 1st column, 1st paragraph).

21. Thus, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to perform the method of toxicological testing and make the recombinant cell of Kliewer and further modify the protein involved in drug metabolism to include other proteins that contribute to drug metabolism such as in chemoresistance such as glucuronosyl transferases, N-acetyltransgerases, p-glyoproteins, gluthathione transgerases and sulfotransferases as per the teachings of Iyer et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Iyer et al. in the instant recombinant cell and screening method because the advantages of studying expression of the instant drug metabolizing enzymes play a key role in cancer therapy. In addition, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to further modify the cells of the method to included those of an isolate human cell line and or various systems other than the liver (lung, gastrointestinal tract and kidney) as per the teachings of Windmill et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Windmill et al. in the instant recombinant cell and screening method because these drug metabolizing enzymes are not limited to one organ system: for example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung" and "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16).

22. Claims 84-115, 117, 129, 130 and 132 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kliewer et al. (*Cell*, 1998; recent IDS) in view of Iyer et al. (*Europ. J. Cancer*, 1998) and Windmill et al. (*Mutation Research*, 1997); and further in view of Makrides (*Protein Expr. Purif.*, 1999).

Kliewer at al demonstrates a screening assay for the induction pregnane X receptor (PXR, a hormone orphan receptor)(*claims 102, 103*) in a transiently transfected isolated mammalian kidney cell line (CV-1; p. 75, 2nd column, 1st paragraph) (*claims 107, 108, 11, 115*). The transfected CV-1 cells "were systematically treated with a series of natural and synthetic compounds that included steroids, vitamin D analogs, thyroid hormone analogs, retinoids, fatty acids, and other small, lipophilic molecules, and reporter levels measured" (p. 75, 2nd column, 1st paragraph) (*claims 84, 99-101*). The cultured recombinant cells (*claim 109*) compromise:

- an extrachromosomal first nucleic acid (*claim 94*) with a human CYP3A1 promoter (a native promoter and also an endogenous promoter) (*claim 84, 97*) a CAT reporter gene (*claims 92, 93*); and
- an extrachromosomal second nucleic acid (*claim 104*) that is also endogenous to the cell (thus within the chromosome of the cell)(*claims 105, 106*) that encodes an PXR which induces the expression of cytochrome P450 proteins involved in drug metabolism (CYP3A1) (*claim 85*),

as required by claim 84 (p. 80, 1st to 2nd column, *Experimental Procedures*). As such Kliewer et al teaches the limitations of the instant claims 84, 85, 92-94, 97, 99-109, 111 and 115.

Kliewer et al does not teach the various other drug metabolizing enzymes of claims 86-91, the various tissues of claims 112-114, the stable transfections of the 1st and/or 2nd nucleic acid (*claims 84, 117*), nor the incorporation of the nucleic acids into a chromosome or endogenous limitations of claims 95, 96, 98, 129, 130 and 132.

Iyer et al. is a teaches the use of pharmacogenetics in screening drug toxicity that evaluate specific drug metabolizing enzymes (*claims 86-91*) including glutathione S-thansferases, uridine diphosphate glucuronosyl-transferases, and cytochrome P450 enzymes (abstract, lines 5-8).

Windmill et al. demonstrates the expression of various drug metabolizing enzymes (*claims 112-114*) in a multitude of human (*claims 110*) organ systems. For example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder, ureter, stomach and lung"; "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16); and a cytochrome P450

expression in "human liver, stomach, small and large intestine, gall bladder, appendix, lung, kidney and adrenals (p.156, 1st column, 1st paragraph).

Makrides teaches the advantages in developing mammalian gene expression vectors that can be successfully transiently or stably transfected for a broad range of cell types (abstract). While Makrides teaches that with respect to transient transfection, "the method lends itself to the rapid testing of vector functionality as well as optimization of different combinations of promoters and other elements in expression vectors" (p.184, 2nd column, 1st paragraph). Makrides also teaches that stable transfections (*claims 84, 117*) are also desirable because the gene expression within the cell lines would be 'permanent' (p. 185, 1st column, 2nd paragraph). In addition, Makrides states that stable transfections can be either a "stable integration of plasmid into the host chromosome [(*claims 95, 96, 105, 129, 130*); it is to be noted, once a cell is stably transfected the transfected nucleic acids become endogenous to the progeny (*claim 98, 132*); OR] it is also possible, however, to generate stable cell lines that harbor vectors extrachromosomally [(*claim 94*)]" (p. 185, 1st column, 2nd paragraph).

23. Thus, it would have been *prima facia* obvious to one of ordinary skill in the art at the time of the invention was made to perform the method of screening and make the recombinant cell of Kliewer, and further modify the protein involved in drug metabolism to include other proteins that contribute to drug metabolism such as in chemoresistance such as glucuronosyl transferases, N-acetyltransgerases, p-glyoproteins, gluthathione transgerases and sulfotransferases as per the teachings of Iyer et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Iyer et al. in the instant recombinant cell and screening method due to the advantages of studying expression of the instant drug metabolizing enzymes play a key role in cancer therapy. In addition, it would have been obvious to one of ordinary skill in the art at the time of the invention was made to further modify the cells of the method to included those of an isolate human cell line and or various systems other than the liver (lung, gastrointestinal tract and kidney) as per the teachings of Windmill et al. Thus, one of ordinary skill in the art would have been motivated to do the modifications taught by Windmill et al. in the instant recombinant cell and screening method due to these drug metabolizing enzymes are not limited to one organ system: for example N-acetyltransferase "mRNA expression was detected in human liver, small intestine, colon, esophagus, bladder,

ureter, stomach and lung" and "sulfotransferase expression in the human colon, small intestine, lung, stomach and liver" (abstract, lines 12-16). Furthermore, it would have been *prima facia* obvious to one of ordinary skill in the art at the time of the invention was made to perform the method of Kliewer in view of Iyer et al. and Windmill et al.; and further stably transfect at least one of the nucleic acids as per the teaching of Makrides. One of ordinary skill in the art would have been motivated to modify the method and recombinant cell because the alternative embodiments allow for greater versatility in the utility of the method and recombinant cell. As per the teachings of Makrides, the nucleic acids can exist stably in various capacities wherein the 1st and/or 2nd nucleic acids can be either stably incorporated into the genome of the cell **or** be stably transfected while remaining as an extrachromosomal element. In addition, an ordinary artisan would have been further motivated to perform the modified method with the first and/or the second nucleic acid molecules stably transfected due to the advantages to having a cell line for a screening assay that permanently expresses the transfected construct instead a cell line with limited timeline of expression as seen in transient transfections.

Conclusion

MAINTAINED REJECTIONS

- Claims 116, 118, 119, 126-131, 133-137, 139-143, 145 and 146 are rejected under 35 U.S.C. 102(b) as being anticipated by Honkakoski et al.
- Claims 120-125, 144 and 147-149 are rejected under 35 U.S.C. 103(a) as being unpatentable over Honkakoski et al. in view of Iyer et al. and Windmill et al.

NEW GROUNDS FOR REJECTIONS

- Claims 84, 85, 92, 93, 95-101, 105-112, 116, 117, 119, 120. 126, 127, 129-135 and 139-146 are rejected, as necessitated by amendment, under 35 U.S.C. 102(b) as being anticipated by Quattrochi et al.
- Claims 116, 118, 119, 126-128, 131, 133-143, 145 and 149 are rejected, as necessitated by amendment, under 35 U.S.C. 102(b) as being anticipated by Kliewer et al. (recent IDS).

- Claims 84-115, 117, 128-130, 132, 138 and 139 are rejected, as necessitated by amendment, under 35 U.S.C. 103(a) as being unpatentable over Honkakoski et al. in view of Iyer et al. and Windmill et al.; and further in view of Makrides.
- Claims 86-91, 113-115, 120-125 and 147-149 are rejected, as necessitated by amendment, under 35 U.S.C. 103(a) as being unpatentable over Quattrochi et al. in view of Iyer et al. and Windmill et al.
- Claims 120-125, 144 and 146-148 are rejected, under 35 U.S.C. 103(a) as being unpatentable over Kliewer et al. (recent IDS) in view of Iyer et al. and Windmill et al.
- Claims 84-115, 117, 129, 130 and 132 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kliewer et al. (recent IDS) in view of Iyer et al. and Windmill et al.; and further in view of Makrides.

No claim is allowed.

Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Inquiries

Papers related to this application may be submitted to Technical Center 1600 by facsimile transmission. Papers should be faxed to Technical Center 1600 via the PTO Fax Center located in Crystal Mall 1. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG

30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993) (See 37 CFR § 1.6(d)). The central **Fax number is (703) 872-9306.**

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Monika B. Sheinberg, whose telephone number is (571) 272-0749. The examiner can normally be reached on Monday-Friday from 9 A.M to 5 P.M. If attempts to reach the examiner by telephone are unsuccessful, the primary examiner in charge of the prosecution of this case, Jehanne Sitton, can be reached at (571) 272-0752. If attempts to reach the examiners are unsuccessful, the examiner's supervisor, Gary Benzion, can be reached at (571) 272-0782.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to Patent Analyst, Chantae Dessau, whose telephone number is (571) 272-0518, or to the Technical Center receptionist whose telephone number is (703) 308-0196.

February 9, 2004
Monika B. Sheinberg
Art Unit 1634
MBS

Jehanne Sitton
Primary Examiner
2/9/04